FINAL REPORT

EFFECTS OF UV-B RADIATION ON SELECTED LEAF PATHOGENIC FUNGI AND ON DISEASE SEVERITY

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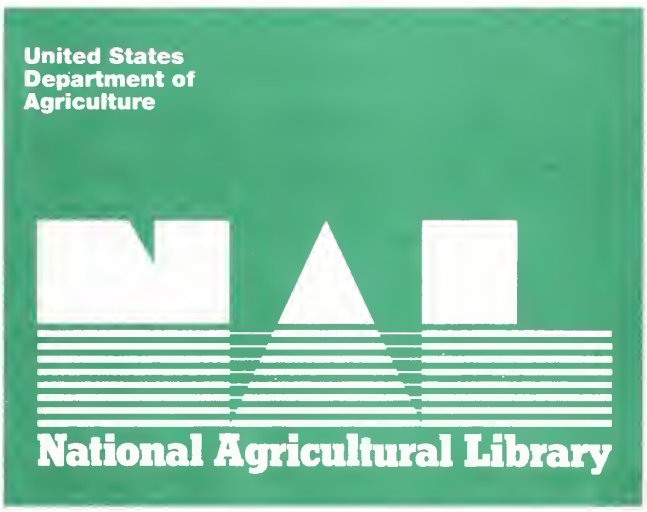
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ABSTRACT

The results of increased levels of UV-B irradiance on spore germination indicate that although plant leaf pathogenic fungal species vary considerably in sensitivity to UV-B, relatively high irradiance levels are required to reduce germination percentage. Pigmented spores such as Cladosporium, Stemphylium, and Alternaria were found to be more resistant to increased UV-B irradiance than hyaline spores (Mycosphaerella, Colletotrichum).

Disease severity of Colletotrichum on cucumber was decreased with increasing UV-B irradiance. A linear decrease in the percentage of leaf area diseased with increased irradiance was found.

Level of UV-B irradiance did not affect severity of disease incited by Cladosporium.

There were no noticeable UV-B effects on either the Stemphylium pathogen or the host, alfalfa. Alternaria, the tomato pathogen, reacted similarly to Cladosporium on cucumber, disease severity being unaffected by UV-B irradiance levels.

Recognizing that our results represent only a small sampling of leaf disease organisms and plant disease-interaction experiments, they appear to support the following: considerably higher levels of UV-B irradiance than those expected from the projected ozone depletion will be required to adversely affect germination and growth of leaf pathogenic fungi; and, where fungal germination and growth are affected, disease severity in the host plant can be expected to be reduced as UV-B irradiance increases.

INTRODUCTION

The impact of increased radiation in the 280-320 nm region (hereafter referred to as UV-B) on overall plant growth, development and critical metabolic processes involved is a major concern in the short-term Biological and Climatic Effects Research Program. Of equal concern is the possibility that crop yield and quality may be affected indirectly by influencing plant susceptibility to various plant pathogens and/or by affecting the pathogens directly.

Numerous studies have been made on the influence of visible light on sporulation of fungi; a limited number of studies have been conducted on the effects on plant infection and disease severity. A few reports have dealt with the effects of ultraviolet radiation; however, most researchers used germicidal lamps (major energy in the 254 nm range) (UV-C). UV-C from germicidal lamps enhanced sporulation of some fungi and inhibited or retarded it in others. UV-C effects on pathogen spore germination, mycelial growth and subsequent infection and disease severity have also been studied and generally an adverse effect on the pathogen has been reported. In addition, limited information is available on the effects of other UV unspecified or poorly defined wavelengths on fungal behavior. Numerous references are cited in reviews by Marsh et al. (1959) and by Leach (1971).

Our research was undertaken to provide preliminary information on the effects of UV-B radiation on spore germination, mycelial growth, infectivity and disease severity of three fungal leaf pathogens of cucumber (Cucumis sativus L.), one of tomato (Lycopersicon esculentum Mill.), and two of alfalfa (Medicago sativa L.). The pathogens selected were: Colletotrichum lagenarium (Pass.) Ell. & Halst., causing cucumber anthracnose;

Cladosporium cucumerinum Ell. & Arth., causing cucumber scab; Mycosphaerella melonis (Pass.) Chiu & J. C. Walker, causing cucumber black rot; Stemphylium botryosum Wallr., causing an alfalfa leafspot; Uromyces striatus Schroet. var medicaginis (Pass.) Arth., causing alfalfa rust; and Alternaria solani (Ell. & Mart.) L. R. Jones & Groot, causing tomato early blight.

MATERIAL AND METHODS

UV-B Radiation - Measurements, Instrumentation, and Methodology

UV-B enhancement facilities were developed cooperatively with the Agricultural Equipment Laboratory (AEL), Beltsville Agricultural Research Center. UV-B enhancement was provided by the required assembly of Westinghouse[1/] FS40 or FS20 fluorescent sunlamps, either filtered with 6-hour-aged 5 mil cellulose acetate (CA) (plus UV-B) or 5 mil Mylar (minus UV-B).

Spore germination and mycelial growth experiments were carried out in temperature-controlled incubators equipped with one FS20 lamp each as the UV-B source. Three incubators were equipped with one FS20 lamp as the only radiation source and one incubator was additionally equipped with two Westinghouse 14-watt cool white fluorescent lamps to provide visible energy.

Greenhouse and growth chamber experiments dealing with pathogen infectivity and disease severity were carried out in a fiberglass greenhouse or in plant growth chambers equipped with FS40 lamp assemblies provided by AEL.

1/ Mention of a trademark, proprietary product, or vendor does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products or vendors that may also be suitable.

UV-B irradiance levels were determined for each plant or fungal location in each experiment with either an Optronics Laboratories, Inc. Model 725 UV-B Radiometer or an Instrumentation Research Laboratory (IRL) UV-B Radiometer described in the IRL final report. Radiometer readings were verified by spectral irradiance determinations (250-369 nm) with an automated spectroradiometer as described in the IRL report at selected locations in the experimental irradiation areas.

Weighted irradiance levels are reported as $mWm^{-2}BUV$, the biologically effective UV derived from the $A\Sigma 9$ weighting function, and unweighted irradiance as mWm^{-2} obtained by summing the measured or calculated values at each nanometer from 280-320 nm. Dividing $mWm^{-2}BUV$ by 3.06 (the $mWm^{-2}BUV$ of control sunshine) provides the fraction of BUV received by each plant or fungal location relative to that of one control sunshine.

Since all UV irradiation for experiments reported here was filtered through cellulose acetate, BUV was limited to the UV-B region (280-320 nm).

For details concerning average control sunshine, spectral characteristics of UV fluorescent lamps and filters, and the weighting function, see the BACER final reports of the Agricultural Equipment Laboratory, and the Instrumentation Research Laboratory, Beltsville Agricultural Research Center.

Spore Production

Sporulating cultures of *Cladosporium cucumerinum* and *Mycosphaerella melonis* were grown on potato dextrose agar at room temperature (22-24°C). *Colletotrichum lagenarium* was maintained under the same conditions on V-8 juice agar. For *Stemphylium botryosum*, sporulation was induced by placing V-8 juice agar cultures in a 22°C incubator fitted with four

Westinghouse 14-watt cool white fluorescent lamps on a 12-12 hour light-dark cycle. Alternaria solani spores were obtained by growing mycelia on lima bean extract agar. After 7 days of culture, the mycelia were scraped with a scalpel in a sterile transfer chamber and the Petri dish lid was removed. Spores were produced the following day. Uredospores of Uromyces striatus var. medicaginis, an obligate parasite, were obtained from infected alfalfa plants.

Spore Germination

From a distilled water suspension of fungal spores, a drop was pipetted onto 2 percent water agar in polyethylene plastic Petri dishes and allowed to dry. As the uredospores of Uromyces striatus do not readily suspend in water, spores were dispersed onto the agar surface by dusting with a sterile camel's hair brush. The lids of the dishes were removed and replaced by filter squares of 5 mil CA, pre-solarized for 6 hours, or 5 mil Mylar as required. Dishes were then transferred to incubators at predetermined positions under the FS20 lamp such that they were subjected to weighted irradiance levels of 6.25 mWm^{-2}BUV (644 mWm^{-2}), 6.69 mWm^{-2}BUV (691 mWm^{-2}), 8.06 mWm^{-2}BUV (8.33 mWm^{-2}) and 10.52 mWm^{-2}BUV (1087 mWm^{-2}).

Samples were irradiated for 6 hours, then left in the dark for 18 hours after which microscopic counts of spore germination (200-500 spores per dish) were made. Each UV-B irradiance was duplicated within each of three incubators. A fourth incubator was additionally supplied with 25 $\mu Em^{-2}s^{-1}$ of photosynthetically active radiation (PAR) provided by cool-white fluorescent lamps. These lamps were allowed to remain on during the 6-hour UV-B irradiation and for an additional 6 hours thereafter.

Mycelial Growth

Fungal cultures were exposed to UV-B in the same incubators used for spore germination. The 5 mil CA filter was placed as a collar around

the FS20 lamp, lids were removed and the Petri dish bottoms were enclosed by UV-B transparent polyethylene bags to keep the agar from drying out over the course of the experiments.

Samples were inoculated by placing a 7 mm diameter core of mycelia upside down in the center of the agar plates which were then placed at the predetermined positions in the chambers. Samples were irradiated daily for 6 hours until growth reached the perimeter of the Petri dish or sufficient data points had been accumulated. Growth was determined by daily measuring the diameter of the colonies.

Disease Development

Cucumbers

For epidemiological experiments with cucumbers, the UV-B sensitive cultivar, Poinsett, was germinated in a synthetic soil mix of peat and vermiculite (Jiffy Mix) in 12.5 cm pots in a fiberglass greenhouse, five seedlings per pot. Temperatures ranged from 24-27°C during the day and 19-21°C at night. Plants were subjected from emergence to UV-B radiation supplied by eight FS40 lamps, filtered by pre-solarized 5 mil CA. Filters were changed every fourth day. Plants were irradiated with UV-B between the hours of 1000 and 1600 daily for the duration of the experiment. Figure 1 shows a typical experimental design used in the plastic greenhouse.

As the cotyledons became fully expanded (5-7 days after seeding), the seedlings were selected for uniformity and thinned to one per pot. Plants were inoculated when the first leaf was fully expanded (11-14 days).

Colletotrichum lagenarium. Figures 2 and 3 diagram the experimental design and provide the weighted and unweighted UV-B irradiances, respectively, for each pot location.

Figure 1. Typical experimental set-up used in UV-B enhancement studies in the greenhouse containing a four-fixture, two lamps per fixture, array of FS40 fluorescent sunlamps (filtered with 5 mil CA). 'Poinsett' cucumber plants infected with Colletotrichum are shown. The typical chlorotic lesion response induced by UV-B irradiation is evident. Disease symptoms are visible at the bottom of the photograph on plants receiving the lowest levels of UV-B irradiance.

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474	631	791	948	1043	1043	1043	982	854	724	599	441	316	221	158	125	98
506	665	823	982	1072	1138	1138	1043	948	791	631	441	349	221	158	125	98
537	696	858	1072	1168	1233	1199	1138	1010	823	665	474	349	221	190	125	98
568	727	948	1138	1233	1296	1263	1168	1043	854	665	474	349	253	190	125	98
537	727	948	1105	1199	1263	1233	1168	1043	854	665	474	349	253	190	125	98
537	696	858	1010	1138	1199	1199	1105	982	823	631	474	349	221	158	125	98
474	631	823	948	1043	1072	1072	1010	858	724	599	441	316	221	158	125	98
441	599	727	854	982	1010	1010	948	823	696	537	380	284	190	158	125	98

Figure 3. Colletotrichum greenhouse experimental arrangement. Each number represents the total unweighted UV-B irradiance in mWm^{-2} at each plant canopy. Plants were 0.15 m apart. Brackets indicate positions of fixtures, with the outer fixtures being 0.8 m, and the inner fixtures 1.0 m from the top of the pot.

An inoculum, consisting of 30,000 spores/ml suspended in distilled water, was prepared from 8-day old Colletotrichum cultures. Using an electric sprayer, the leaves were covered with fine droplets of inoculum. Control plants were sprayed with distilled water. To attain the high relative humidity necessary for infection, the sprayed plants were enclosed within UV-B transmittable polyethylene bags for a 48-hour inoculation period.

Photographs, fresh weight, dry weight, area of first leaf, and percent of first leaf diseased were recorded and used to determine UV-B-disease interaction.

Cladosporium cucumerinum. The weighted and unweighted UV-B irradiances for each pot location are shown in Figures 4 and 5, respectively. The inoculum, containing 70,000 spores/ml, was applied as described for Colletotrichum and the inoculated plants were sealed in polyethylene bags for 48 hours. Plants were harvested 8 days after inoculation. Fresh weight, dry weight, and area of first leaf were used to determine UV-B-disease interaction as disease symptoms did not permit precise scoring.

Mycosphaerella melonis. The weighted and unweighted UV-B irradiances, respectively, for each pot location are shown in Figures 6 and 7. The inoculum, containing 60,000 spores/ml, was applied as for Colletotrichum. Similar data were also taken; however, as with Cladosporium, disease symptoms did not permit precise scoring.

Alfalfa

For the evaluation of the effect of UV-B on leaf rust of alfalfa, the cultivar Arc was grown in a greenhouse for 5 weeks and then transferred to two plant growth chambers. Temperature in the chambers was 25-20°C day-night with a relative humidity of 90 percent. An average of 200 $\mu Em^{-2}s^{-1}$

4.9	6.7	9.2	10.7	11.9	12.9	12.9	11.9	10.4	8.9	7.0	5.2	3.7	2.8	1.8	1.2	0.9
5.2	7.7	9.5	11.9	13.2	14.4	14.4	13.5	11.6	9.5	7.7	5.8	4.3	2.8	1.8	1.2	0.9
6.1	8.3	10.7	12.9	14.4	15.0	15.3	14.4	12.6	10.4	8.0	6.1	4.6	3.1	2.1		0.9
6.7	8.6	11.3	13.5	15.0	15.9	15.9	15.0	13.8	10.7	8.3	6.1	4.6	3.4	2.1		0.9
6.7	9.2	11.6	13.5	15.0	15.9	15.6	14.7	12.9	10.4	8.3	6.1	4.3	3.1	2.1	1.2	1.2
6.7	8.6	11.0	12.9	14.4	15.0	15.0	13.5	11.9	9.8	7.7	5.5	3.7	2.8	1.8	1.2	0.9
6.1	8.0	10.1	11.9	12.9	13.5	13.5	12.6	11.0	9.2	7.0	5.2	3.7	2.8	1.8	1.2	0.9
5.8	7.0	9.5	11.3	12.2	12.9	12.6										

Figure 4. Cladosporium greenhouse experiment arrangement. Each number represents the biologically effective UV-B irradiance in mWm^{-2} BUV at each plant canopy. Plants were 0.15 m apart. Brackets indicate position of fixtures, with the outer fixture being 0.8 m, and the inner fixture 1.0 m from the top of the pot.

505	696	948	1105	1233	1328	1328	1233	1072	915	727	537	380	284	190	125	95
537	792	981	1233	1358	1484	1484	1389	1199	981	792	599	441	284	190	125	95
631	854	1105	1328	1484	1547	1579	1484	1235	1072	823	631	474	316	234		95
696	885	1168	1389	1547	1641	1641	1547	1421	1105	854	631	474	349	234		95
696	948	1199	1389	1547	1641	1611	1515	1328	1072	854	631	441	316	234	125	125
696	885	1138	1328	1484	1547	1547	1389	1233	1010	792	568	380	284	190	125	95
631	823	1043	1233	1328	1389	1389	1235	1138	948	727	537	380	284	190	125	95
599	727	981	1168	1263	1328	1235										

Figure 5. *Cladosporium* greenhouse experiment arrangement. Each number represents the total unweighted UV-B irradiance in mWm^{-2} at each plant canopy. Plants were 0.15 m apart. Brackets indicate position of fixtures, with the outer fixture being 0.8 m, and the inner fixture 1.0 m from the top of the pot.

4.9	6.4	8.7	10.1	11.3	11.9	11.9	11.6	10.4	7.7	6.1	4.6	3.7	2.8	1.8	1.2	.9
5.2	7.0	9.2	11.0	12.2	12.9	12.9	12.2	11.0	8.0	6.4	4.9	3.7	2.8	1.8	1.5	.9
5.8	7.7	9.8	11.6	13.2	14.1	14.1	13.5	11.9	8.7	7.0	6.1	4.0	3.1	2.1	1.5	1.2
6.4	8.7	10.7	12.9	14.4	15.0	15.0	14.4	12.6	10.1	8.3	5.8	4.3	3.1	2.1	1.5	1.2
6.4	8.7	10.4	12.9	14.4	15.0	15.0	14.4	12.6	10.4	8.3	5.8	4.3	3.1	2.1	1.5	.9
6.1	8.0	10.1	12.6	14.1	14.7	14.7	13.8	11.9	9.8	8.0	5.8	4.0	2.8	1.8	1.2	.9
5.8	7.7	9.5	11.6	12.9	13.5	13.5	12.6	11.3	9.2	7.3	5.2	3.7	2.8	1.8	1.2	.9
5.2	6.7	8.1	10.4	11.6	11.9	11.9	11.3	9.8	8.3	6.7	4.6	3.4	2.1	1.8	1.2	.9

Figure 6. Mycosphaerella greenhouse experiment arrangement. Each number represents the biologically effective UV-B irradiance in $mWm^{-2}BUV$ at each plant canopy. Plants were 0.15 m apart. Brackets indicate position of fixtures, with the outer fixture being 0.8 m, and the inner fixture 1.0 m from the top of the pot.

506	661	900	1043	1166	1227	1227	1197	1072	796	628	466	381	291	186	124	93
537	722	949	1134	1259	1331	1331	1259	1134	827	661	506	381	291	186	154	93
598	796	910	1197	1520	1455	1455	1394	1227	900	722	628	414	320	217	154	124
661	900	1104	1331	1485	1547	1547	1485	1301	1043	858	598	444	320	217	154	124
661	900	1072	1331	1485	1547	1547	1485	1301	1072	858	598	444	320	217	154	93
628	827	1043	1301	1455	1518	1518	1425	1227	910	827	598	414	291	186	124	93
598	796	981	1197	1331	1394	1394	1301	1166	949	753	537	381	291	186	124	93
537	692	835	1072	1197	1227	1227	1166	910	858	691	466	352	217	186	124	93

Figure 7. Mycosphaerella greenhouse experiment arrangement. Each number represents the total unweighted UV-B irradiance in mWm^{-2} at each plant canopy. Plants were 0.15 m apart. Brackets indicate position of fixtures, with the outer fixture being 0.8 m, and the inner fixture 1.0 m from the top of the pot.

visible radiation was provided by sixteen 165-watt cool white fluorescent lamps and twelve 50-watt incandescent bulbs with a photoperiod of 16-hour day 8-hour night. The plants were allowed to acclimate to chamber conditions for 5 days before being irradiated with UV-B. UV-B radiation was provided by two FS40 Westinghouse sunlamps (with no reflector) filtered by 5 mil CA that had been pre-solarized for 6 hours. Filters were changed every fourth day. Figures 8 and 9 diagram the plant arrangement within the chambers and show the weighted and the unweighted UV-B irradiances, respectively, at the canopy height of each plant. Plants were clipped during the experiment to maintain a distance of 0.37 m from the fixture.

Uromyces striatus. The plants were inoculated with spores of the rust fungus after 7 days of UV-B irradiation. Inoculum was prepared by scraping the spores from 10-day-old S. botryosum agar cultures, suspending them in distilled water and filtering the mixture through cheesecloth to remove mycelial fragments. The resulting spore suspension contained 10,000 spores/ml and was applied to the alfalfa leaves in fine droplets by means of a chromatography sprayer. Control plants were sprayed with distilled water only. All plants were then covered for 24 hours with UV-B transmittable polyethylene bags to achieve maximum humidity necessary for good infection. Five days after the end of the inoculation period, all leaflets in the upper 4 centimeters of the plant were scored for type of lesion as follows: 1 = pinhead size brown flecks; 2 = lesion approximately 1 mm in diameter with a brown margin and tan center; 3 = lesion approximately two to three times larger than #2 type and often with obvious yellow halo outside of the brown margin; and 4 = a large blighted area most often found on leaflet margin.

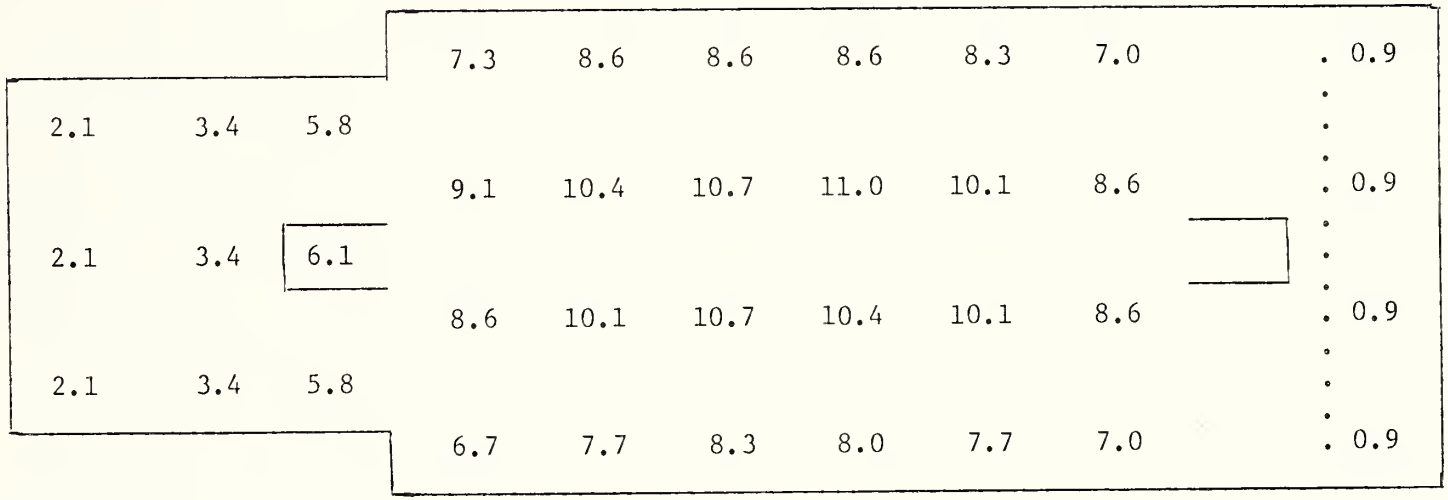

Figure 8. Growth chamber experimental arrangement. Each number represents the biologically effective UV-B irradiance in mWm^{-2}BUV at each plant canopy (0.37 m from the fixture). Plants were 0.15 m apart. Brackets indicate the position of the fixture. The dotted line represents a 5 mil Mylar barrier.

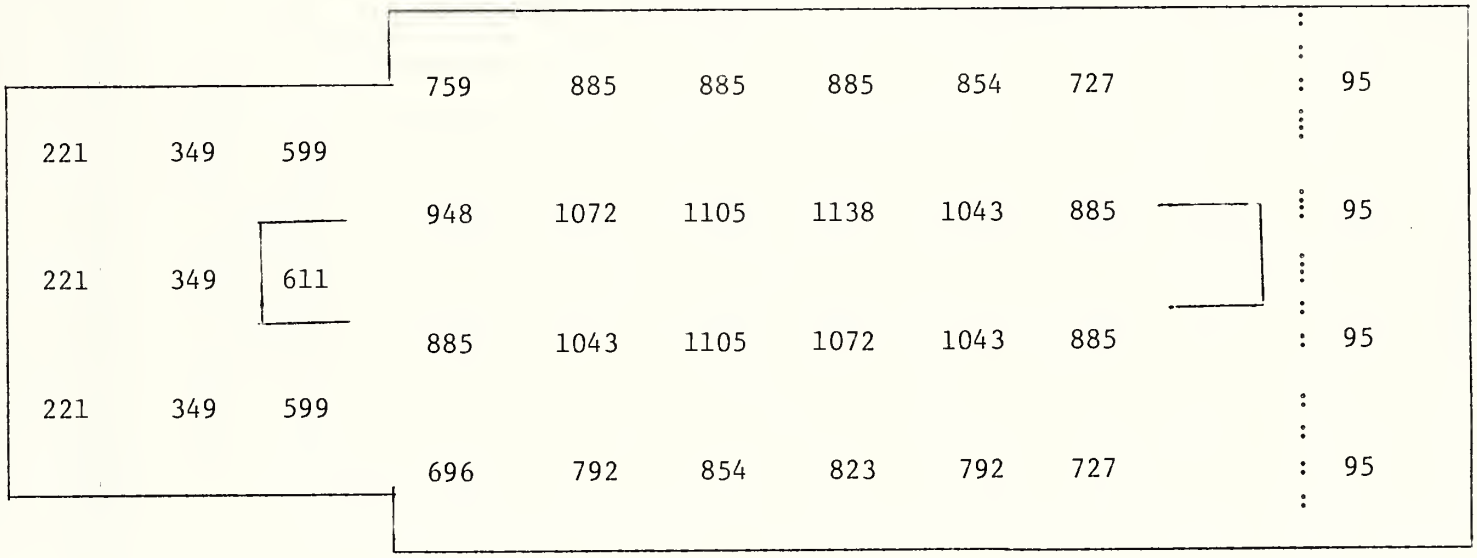

Figure 9. Growth chamber experimental arrangement. Each number represents the total unweighted UV-B irradiance in mWm^{-2} at each plant canopy (0.37 m from the fixture). Plants were 0.15 m apart. Brackets indicate the position of the fixture. The dotted line represents a 5 mil Mylar barrier.

Tomato

The cultivar Chef was germinated and grown in the same plant growth chambers used for the alfalfa experiments except that the FS40 lamps were repositioned to adjust UV-B irradiance levels and temperatures were maintained at 26°C day - 20°C night. Figures 10 and 11 diagram the plant arrangement within the chamber and list the weighted and unweighted irradiances, respectively.

Alternaria solani. Twenty-two days after seeding when the third leaf was well expanded, buds were pinched out; and four days later the plants were inoculated as follows: The spore suspension was prepared by placing two spore mats and 100 ml distilled water in a blender for 15 seconds and then filtering the resulting suspension through cheesecloth to remove mycelia and agar fragments. The suspension was applied in a fine mist to the second and third leaves by means of a chromatography sprayer. Relative humidity in the chamber was maintained at 90-100 percent for 24 hours by a chamber humidity regulator and the placement of a polyethylene canopy over the plants. The second and third leaves were harvested for dry weights 48 hours after inoculation when blighting became severe.

For analyses of plant-disease response as a function of UV-B, in the above experiments, data from individual plants were combined into irradiance groups of increasing 1.5 mWm^{-2}BUV and subjected to analyses of variance. If P = 0.05 or less, the data were further analyzed using linear regression.

RESULTS AND DISCUSSION

Spore Germination

The results of increased levels of UV-B irradiance on spore germination are shown in Table 1. They indicate that even in the more sensitive species, high UV-B irradiance levels are required to reduce germination percentage; generally, more than double the UV-B of one control sunshine

1.8	2.5	3.7	4.6	5.5	6.1	6.7	7.0	7.0	6.7	5.5	4.6	3.1	1.8
1.5	2.8	4.6	5.8	7.0	7.7	8.3	8.6	8.6	8.3	7.3	5.5	3.4	1.8
1.5	2.8	4.9	6.7	8.3	8.9	9.5	9.8	10.1	9.5	8.3	6.4	3.7	1.8
1.2	2.8	4.9	7.0	8.3	9.2	9.5	9.8	10.1	9.5	8.3	6.1	3.7	1.8
1.5	2.8	4.6	6.4	7.7	8.3	8.9	8.9	9.2	8.6	7.3	5.5	3.7	1.8
1.5	2.5	3.7	5.2	6.1	6.7	7.0	7.3	7.0	6.7	5.8	4.3	3.1	1.8

Figure 10. Alternaria growth chamber experimental arrangement. Each number represents the biologically effective UV-B irradiance in $mWm^{-2}BUV$ at each plant canopy.

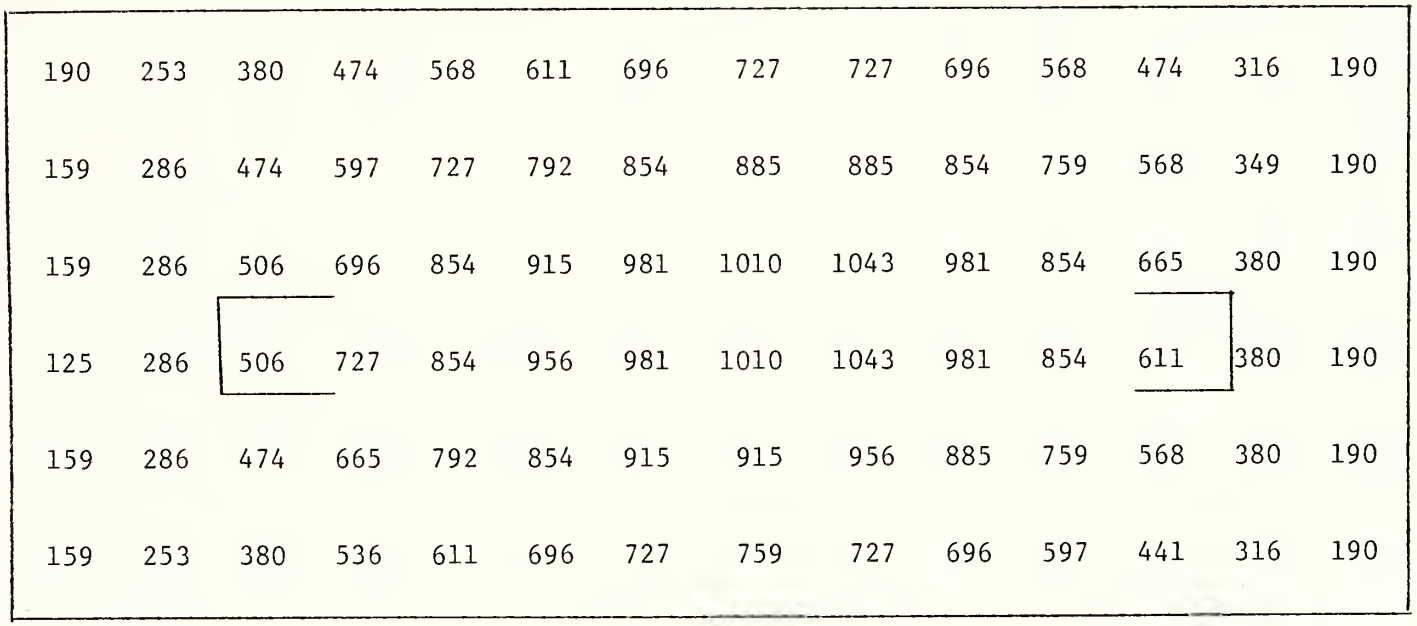

Figure 11. Alternaria growth chamber experimental arrangement. Each number represents the total unweighted UV-B irradiance in mWm^{-2} at each plant canopy.

was required before a reduction in spore germination was noted. In the more resistant species, more than three times the level of control sunshine UV-B was needed to inhibit spore germination.

For Colletotrichum, the most sensitive species, spore germination was reduced by less than two times control sunshine as indicated by comparison of the Mylar-filtered controls and the lowest irradiance level used.

Resistance to UV-B appeared to be correlated with spore pigmentation. Spores of the resistant species Cladosporium, Stemphylium, Uromyces, and Alternaria are all darkly pigmented, whereas, spores of Mycosphaerella and Colletotrichum are hyaline. Our results suggest that pigmentation provided protection from damage by UV-B.

In the incubator supplied with PAR in addition to UV-B, germination values were consistently, but only slightly, higher than those observed in the incubators irradiated by UV-B only, with the most notable increase in the sensitive Colletotrichum spores. Furthermore, we observed that germ tube length appeared to be considerably increased in nearly all tests in the presence of PAR. This observation is consistent with other BACER research which provides evidence for the existence of a photorepair or photoprotection mechanism. Linear regression analyses indicate that within the range of irradiance levels tested, there is a significant correlation between reduction in germination and increased UV-B irradiance in the susceptible species.

Mycelial Growth

A measure of the mycelial growth rate under identical UV-B irradiances and environment used for the spore germination experiments is presented in Table 2. Growth rate is expressed as increase in colony diameter with time. In contrast to the spore germination, increase in colony diameter (as expressed in terms of percentage of the Mylar control) at the end of the

Table 1. Influence of UV-B irradiation on spore germination of six pathogens. Spores were irradiated in Petri dishes in an incubator at 22°C under an FS20 fluorescent sunlamp filtered with either 5 mil cellulose acetate or 5 mil Mylar.

	$mWm^{-2}BUV$					
	Mylar-5 mil	Cellulose acetate-5 mil				
Disease organism	< 0.25	6.26	6.69	8.06	10.52	
	Mean percent germination					r^2
Colletotrichum lagenarium						
Test A	17.82	12.70	17.79	5.13	2.03	0.691
B	49.29	10.31	8.20	2.80	1.22	0.851
Mycosphaerella melonis						
Test A	52.46	57.75	57.34	47.85	35.44	0.945
B	50.21	45.48	42.34	30.95	22.94	0.934
C	41.30	47.85	46.34	37.96	26.62	0.924
D	33.06	29.10	28.49	24.59	19.62	0.885
E	63.08	60.04	57.41	47.28	33.90	0.935
Alternaria solani						
Test A	98.17	98.58	98.49	98.72	98.75	-
B	98.88	99.01	99.38	99.14	99.01	-
Stemphylium botryosum						
Test A	98.08	97.44	97.35	95.49	94.09	-
B	93.46	93.48	93.48	93.04	93.29	-
Cladosporium cucumerinum						
Test A	93.69	90.53	89.02	86.37	81.33	0.942
B	94.73	90.93	89.65	87.80	83.78	0.909
Uromyces striatus						
Test A	86.85	86.70	85.54	86.25	86.10	-
B	90.24	89.62	89.90	89.03	89.88	-

Table 2. Influence of UV-B irradiation on mycelial growth of five pathogens. Mycelia were irradiated in Petri dishes in an incubator at 22°C under an FS20 fluorescent sunlamp filtered with either 5 mil cellulose acetate or 5 mil Mylar.

		$mWm^{-2}BUV$				
		Mylar-5 mil	Cellulose acetate-5 mil			
Disease organism		< 0.25	6.25	6.69	8.06	10.52
	No. days irradiated	Colony diameter - mm				
Colletotrichum lagenarium						
	4	22.8	17.5	15.5	14.0	12.8
	6	34.0	27.0	26.0	26.3	24.0
	8	40.5	35.3	35.0	35.8	29.3
Percent of Mylar Control			87	86	88	72
Mycosphaerella melonis						
	2	45.0	44.0	44.5	43.2	42.7
	3	64.0	62.0	62.7	60.7	60.2
	4	82.3	79.7	80.8	78.3	76.2
Percent of Mylar Control			97	98	95	93
Alternaria solani						
	2	29.8	29.3	29.3	28.8	28.7
	3	40.0	39.0	38.7	38.8	37.5
	4	50.3	49.7	49.3	48.8	48.5
	5	60.5	60.7	59.5	59.0	59.3
	6	71.2	70.7	70.2	69.7	69.7
Percent of Mylar Control			99	99	98	98
Stemphylium botryosum						
	3	28.7	14.8	14.0	13.5	13.3
	5	41.5	23.2	21.7	22.3	18.3
	7	55.7	41.0	39.3	37.7	33.3
	11	78.0	72.5	68.3	68.3	65.0
Percent of Mylar Control			93	88	88	83
Cladosporium cucumerinum						
	3	18.2	12.7	14.0	13.8	13.2
	5	32.9	25.0	24.8	23.7	23.3
	7	47.5	37.2	36.0	34.2	34.3
	11	76.0	59.5	58.5	57.7	55.5
Percent of Mylar Control			78	77	76	73

growth period did not differ greatly from colonies receiving no UV-B. As with spore germination, Colletotrichum appeared to be most sensitive to relatively high UV-B irradiance levels used. Cladosporium and Stemphylium were intermediate in response, while Alternaria and Mycosphaerella showed little if any reduction at the highest UV-B irradiance level used. However, with all species, mycelial density was visibly reduced when compared to the Mylar controls. Attempts to obtain dry weights of colonies revealed differences between CA-filtered and Mylar-filtered colonies, but the method used was insufficiently sensitive to distinguish between CA-treatments. We conclude that, as with spore germination, relatively high UV-B irradiance is required before the growth of these fungi is impaired.

Cucumber

Growth and disease responses of cucumber to UV-B radiation are shown as follows: Colletotrichum, Figures 12 through 17; Mycosphaerella, Figures 18 through 20; and Cladosporium, Figures 21 through 23. Growth responses of the uninoculated control plants, as measured by fresh weight, dry weight, and area of first leaf, responded similarly in all cucumber disease experiments, showing increased repression with each increase in UV-B irradiation level applied.

Disease severity of Colletotrichum and Mycosphaerella on cucumber decreased with increasing UV-B irradiances. This is shown graphically for Colletotrichum (Figure 12) for which percentage of diseased leaf area is plotted against UV-B irradiance levels. Pictorial representation of the response is presented in Figures 16 and 17. Figure 16 depicts the disease response in the absence of UV-B enhancement (Mylar control). In Figure 17, disease response is compared to uninoculated controls subjected to high and low levels of UV-B irradiances. The differences in disease response are obvious.

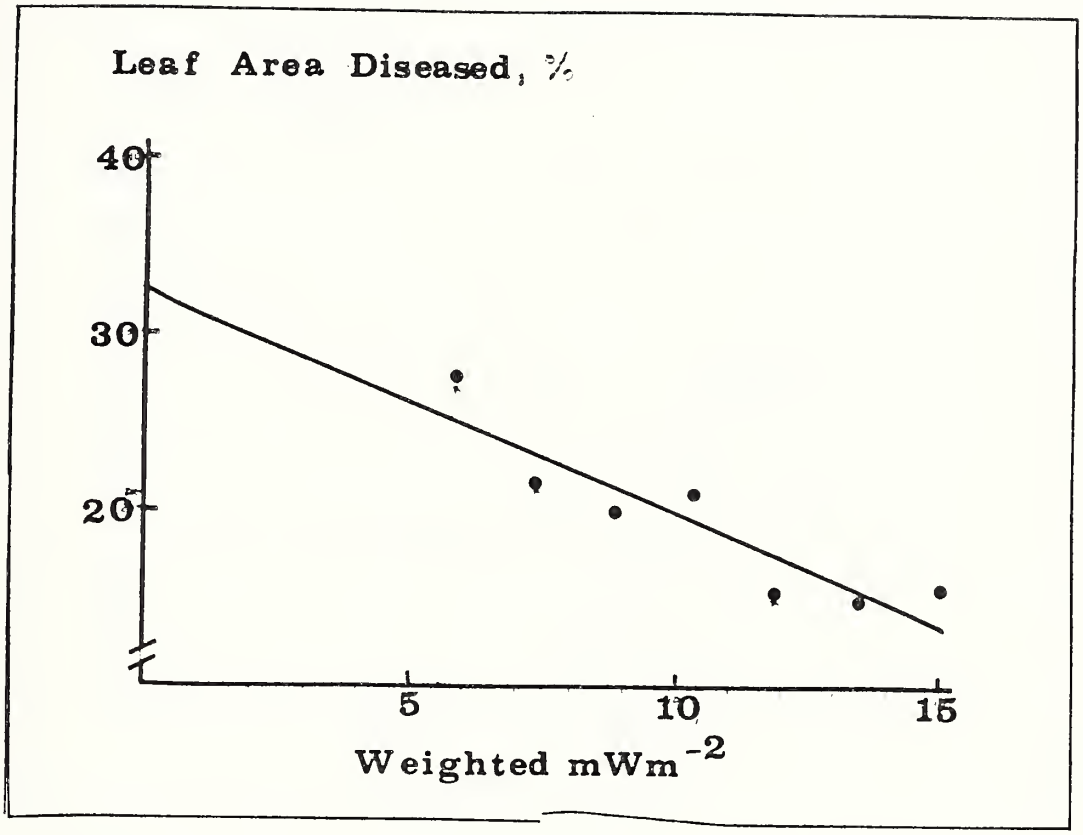

Figure 12. The effect of increased UV-B irradiance on the percent diseased area of cucumber leaves infected with Colletotrichum lagenarium. r^2 = 0.81, standard error = 2.2.

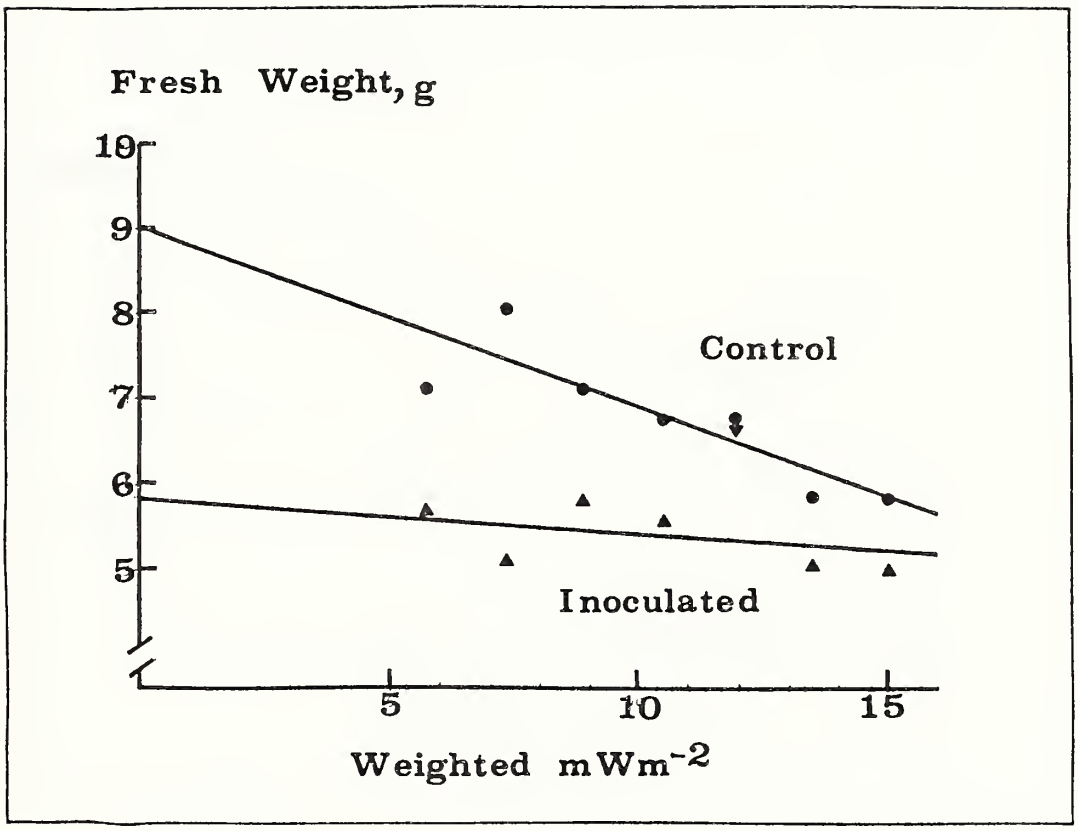

Figure 13. The effect of increased UV-B irradiance on fresh weight of Colletotrichum lagenarium infected (r^2 = 0.77, standard error = 0.63) and noninfected (r^2 = 0.03, standard error = 0.43) cucumber plants.

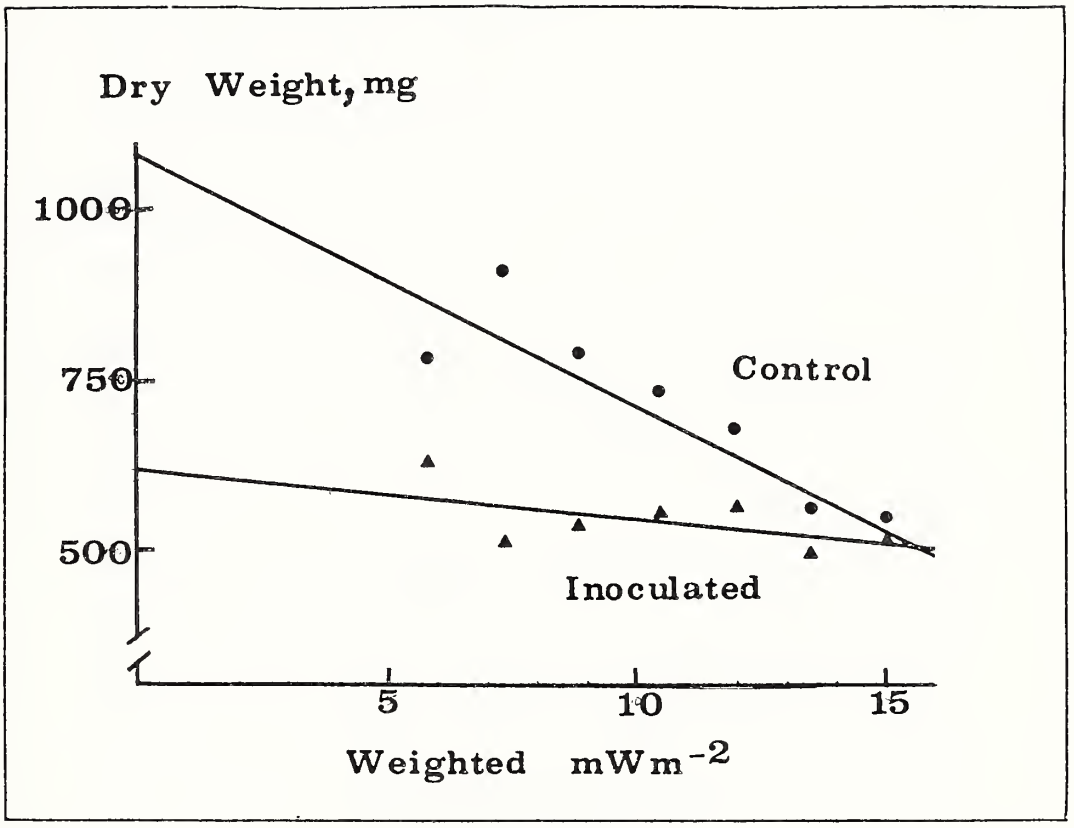

Figure 14. The effect of UV-B irradiance on dry weight of Colletotrichum lagenarium infected (r^2 = 0.32, standard error = 40.6) and noninfected (r^2 = 0.79, standard error = 64.43) cucumber plants.

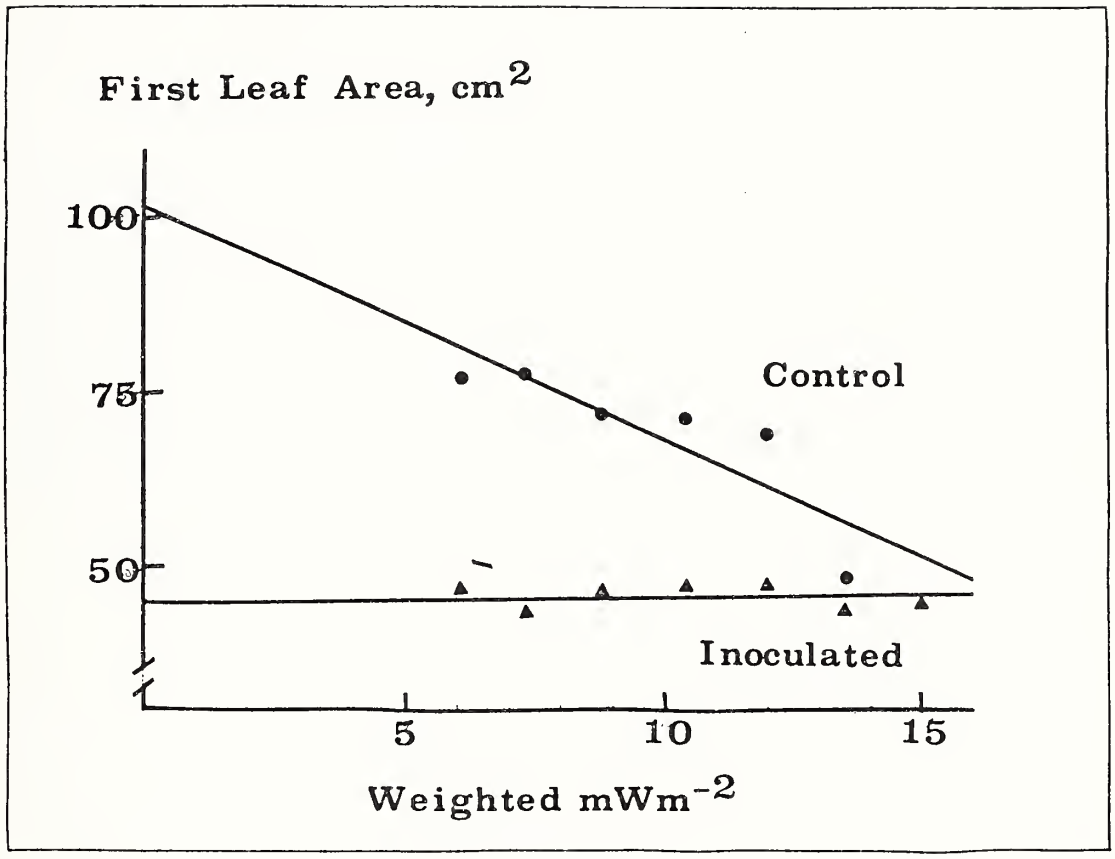

Figure 15. The effect of UV-B irradiance on area of first leaf of Colletotrichum lagenarium infected (r^2 = 0.01, standard error = 2.36) and noninfected (r^2 = 0.80, standard error = 5.74) cucumber plants.

Figure 16. Disease response to inoculation of cucumber with Colletotrichum spores in the absence of UV-B (Mylar filtered); left to right, uninoculated and inoculated plants.

Figure 17. Disease response to inoculation of cucumber plants with Colletotrichum spores subjected to high and low level UV-B irradiance. Left to right: high UV-B uninoculated; high UV-B inoculated; low UV-B uninoculated; and low UV-B inoculated.

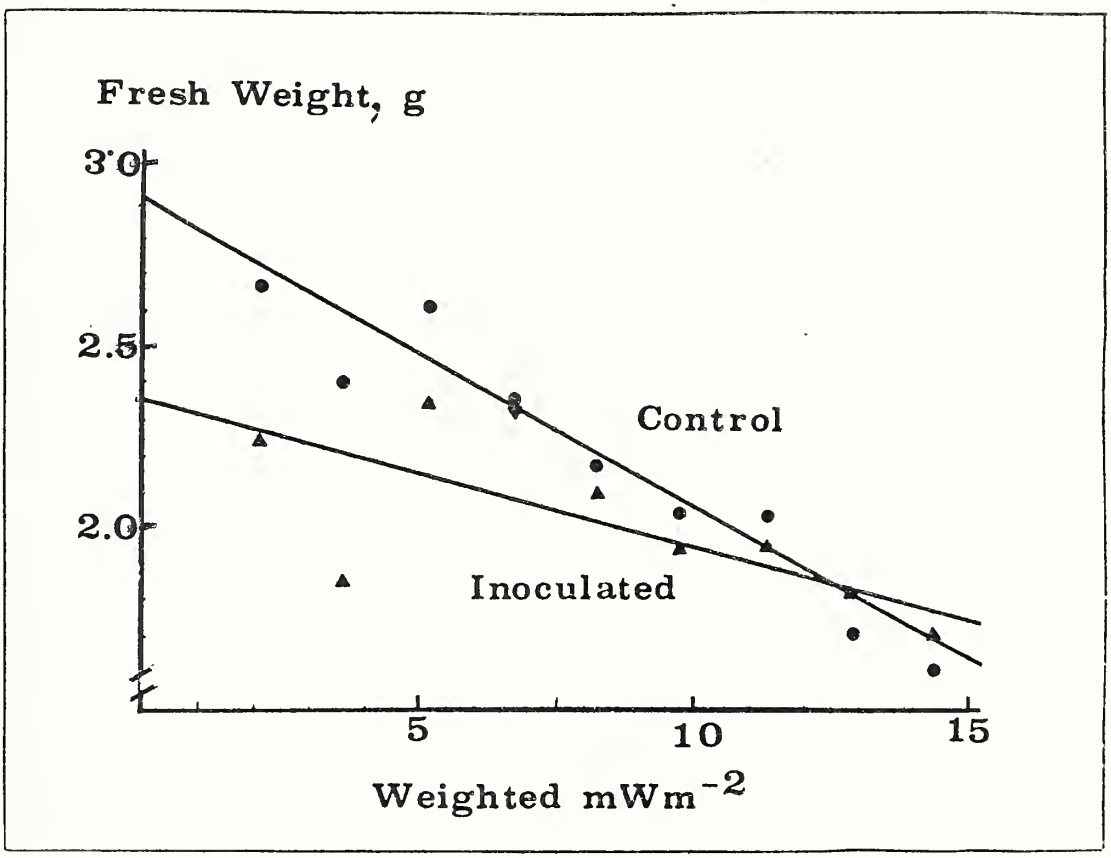

Figure 18. The effect of increased UV-B irradiance on fresh weight of Mycosphaerella melonis, infected (r^2 = 0.43, standard error = 0.19) and noninfected (r^2 = 0.91, standard error = 0.11) cucumber plants.

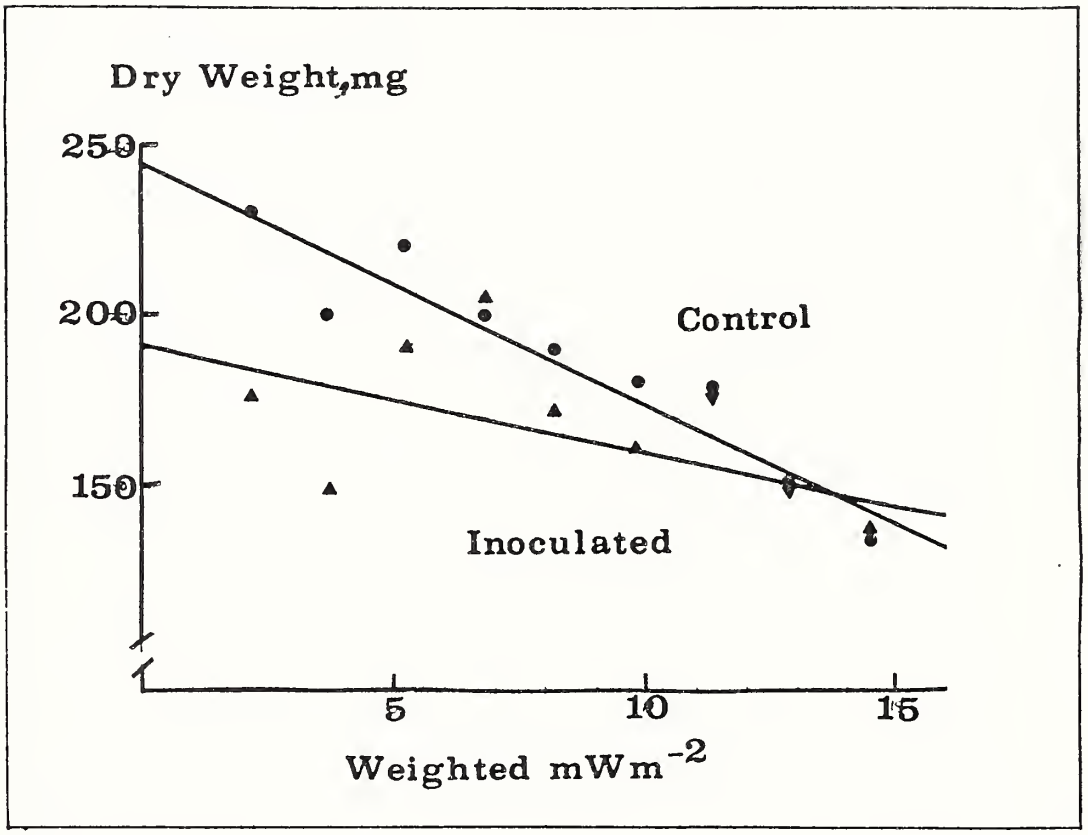

Figure 19. The effect of increased UV-B irradiance on dry weight of Mycosphaerella melonis infected (r^2 = 0.24, standard error = 20.1) and noninfected (r^2 = 0.89, standard error = 10.9) cucumber plants.

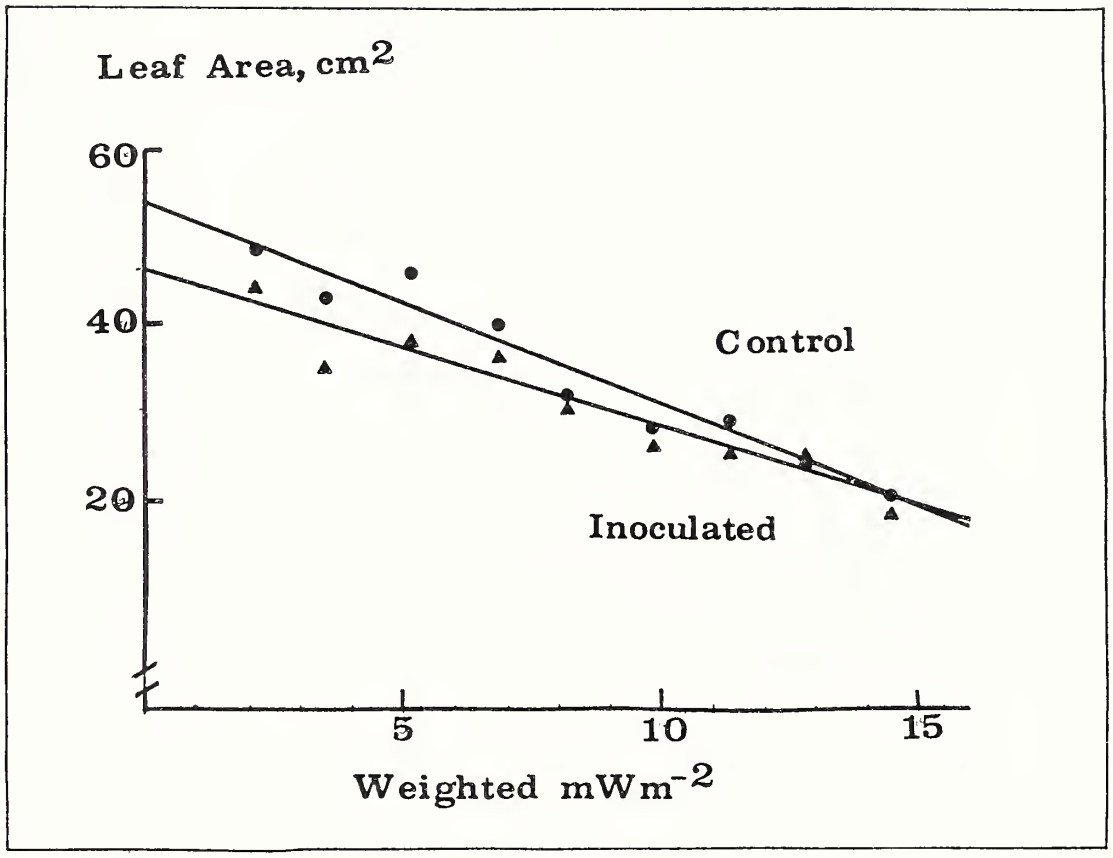

Figure 20. The effect of increased UV-B irradiance on area of first leaf of Mycosphaerella melonis infected (r^2 = 0.91, standard error = 2.61) and noninfected (r^2 = 0.95, standard error = 2.31) cucumber plants.

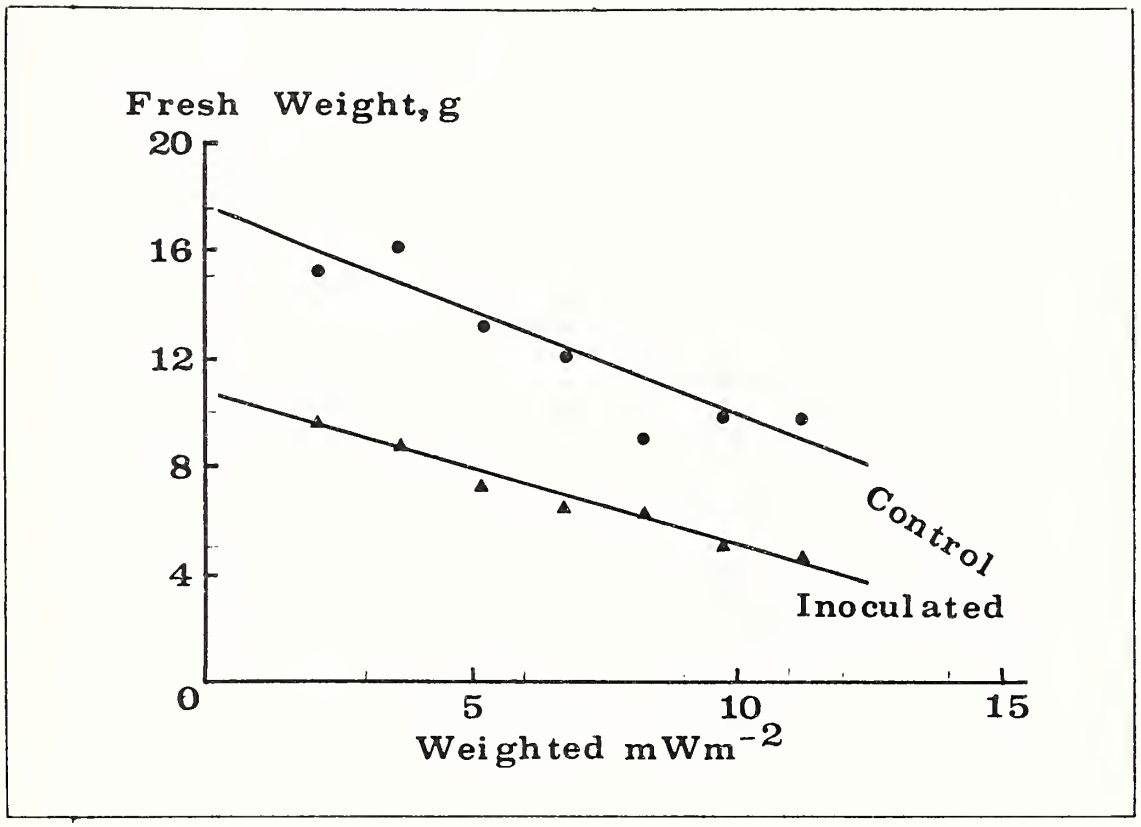

Figure 21. The effect of increased UV-B irradiance fresh weight of Cladosporium cucumerinum infected (r^2 = 0.97, standard error = 0.36) and noninfected (r^2 = 0.84, standard error = 1.24) cucumber plants.

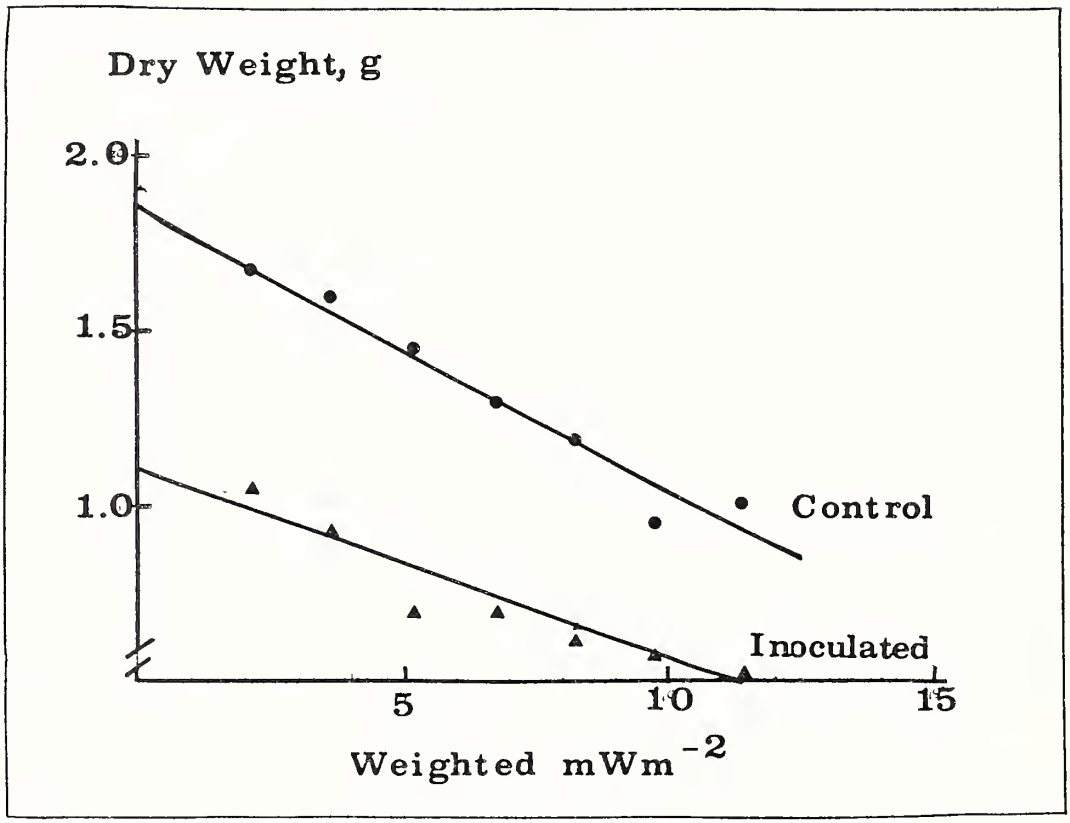

Figure 22. The effect of increased UV-B irradiance on dry weight of Cladosporium cucumerinum infected (r^2 = 0.89, standard error = 0.07) and noninfected (r^2 = 0.94, standard error = 0.07) cucumber plants.

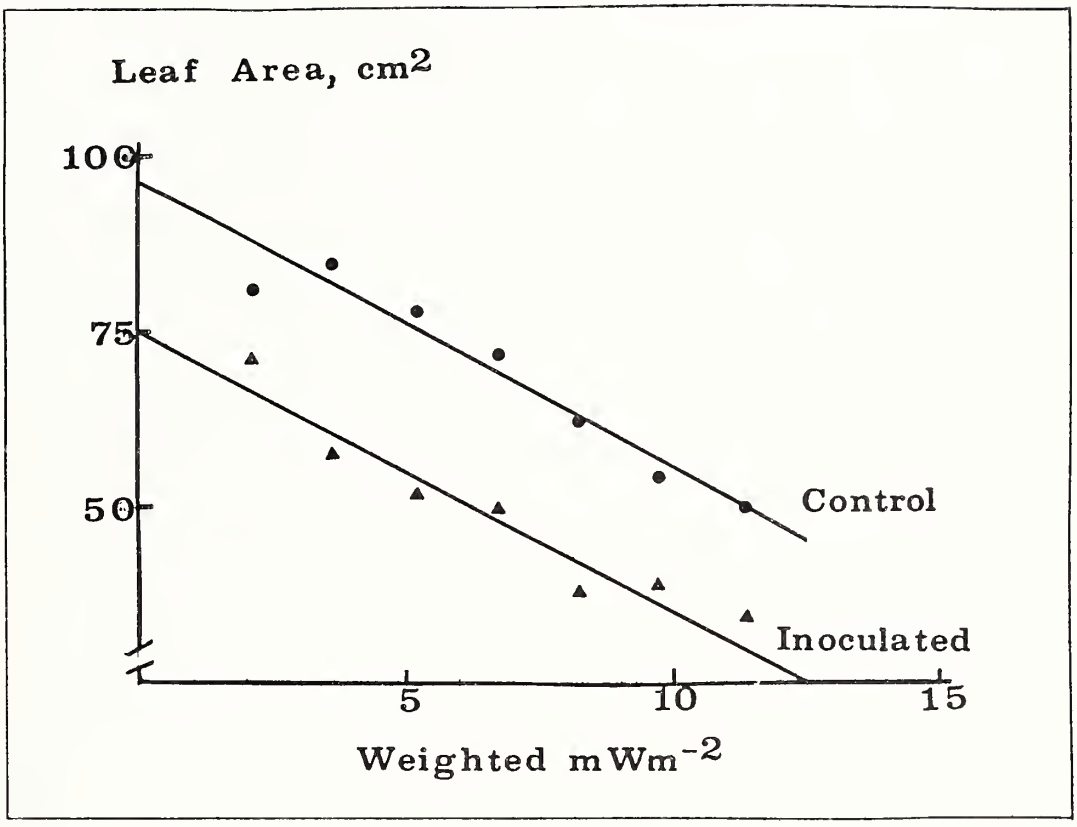

Figure 23. The effect of increased UV-B irradiance on area of first leaf of Cladosporium cucumerinum infected (r^2 = 0.92, standard error = 4.0) and noninfected (r^2 = 0.92, standard error = 4.06) cucumber plants.

The decrease in disease infectivity and severity are reflected in area of first leaf and fresh and dry weight of both Colletotrichum and Mycosphaerella. With both pathogens, the measured growth responses of inoculated and noninoculated plants approach unity in the region of 15 mWm^{-2}BUV irradiance. Colletotrichum and Mycosphaerella both have hyaline spores. These disease-UV-B interactions are consistent with the spore germination and mycelial growth data presented earlier. They suggest that disease response to these organisms is due to the direct effect of UV-B irradiance on the pathogen, although increased resistance to the pathogens by the irradiated cucumber plants could also be a factor.

Cladosporium, possessing a pigmented spore, showed no such pathogen-UV-B interaction. Observed disease symptomology and measurement of first leaf area, fresh and dry weight, showed a uniformly reduced growth of inoculated plants regardless of UV-B irradiance levels (Figures 21 through 23); again, this observation is consistent with the effects noted on spore germination and mycelial growth.

Alfalfa

At the UV-B irradiances used, there were no noticeable UV-B effects on either the Stemphylium pathogen or the host plant, alfalfa. Stemphylium spores are also pigmented. Table 3 shows the results of these experiments undertaken in plant growth chambers and indicates that similar lesion types occurred at all UV-B irradiance levels. The differences between experiments were due to harvesting the first experiment 3 days later than the second. The results indicate that the lesions progressed equally over time under all UV-B levels. Again, this response is consistent with that of other pathogens having pigmented spores. Using Uromyces in an experiment with alfalfa grown in a fiberglass greenhouse, similar results were obtained and supported our

Table 3. The effect of UV-B on disease development of the alfalfa pathogen, Stemphylium botryosum Wallr. on Arc cultivar of alfalfa.

Biologically effective UV-B $mWm^{-2}BUV$	No. leaflets/score	Lesion score			
		Experiment 1		Experiment 2	
		Growth chamber A	Growth chamber B	Growth chamber A	Growth chamber B
2.14	48	2	1	1	1
3.67	48	2	1	2	2
5.20	48	2	2	1	1
6.73	108	1	2	1	1
8.26	240	2	2	1	1
9.76	96	2	1	1	1

belief that the progression of rust on alfalfa is not likely to be affected by enhanced UV-B, except perhaps at very high irradiance levels.

Tomato

Alternaria, a pigmented spore, reacted similarly to Cladosporium on cucumber, disease severity being unaffected by UV-B irradiance levels (Table 4). The differences in dry weight between UV-B irradiances appear to be more closely correlated with PAR than with UV-B radiation.

SUMMARY

Recognizing that our results represent only a small sampling of leaf disease organisms and plant disease interactions, they appear to support the following: 1) considerably higher levels of UV-B irradiances than those expected from projected decrease in ozone that might be caused by chlorofluoromethanes will be required to adversely affect germination and growth of leaf pathogenic fungi, and 2) where fungal germination and growth are affected, disease severity in the host plant can be expected to be reduced as UV-B irradiance increases.

Table 4. The effect of UV-B on disease development of the tomato pathogen, Alternaria solani (Ell. & Mart.) L. R. Jones and Groot.

UV-B weighted $mWm^{-2}BUV$		Dry weight - mg	
		Inoculated	Uninoculated
2.5		148.5	177.3
4.0		168.1	192.0
5.5		190.6	211.4
7.0		172.6	202.9
8.5		198.1	195.6
10.0		131.8	162.5
	Mean	168.3	190.3
	r^2	< 0.01	0.05

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Marsh, Paul B., E. E. Taylor, and L. M. Bassler. A guide to the literature on certain effects of light on fungi: Reproduction, morphology, pigmentation, and phototropic phenomena. Plant Disease Reporter. Supplement 261. U.S. Department of Agriculture. pp 251-312, 1959.